

Applicants : Philip Livingston and Friedhelm Helling
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sphingosine base and (ii) an immunogenic protein-based carrier;

b) a saponin derivable from the bark of a Quillaja saponaria Molina tree; and

c) a pharmaceutically acceptable carrier;

the relative amounts of such conjugate and such saponin being effective to stimulate or enhance production in a subject of an antibody to whichever ganglioside is present as a derivative in the conjugate,

wherein the ganglioside derivative is a derivative of a ganglioside selected from the group consisting of GD2, GD3 lactone, O-acetyl GD3 and GT3;

wherein the immunogenic protein-based carrier is derived from a protein selected from the group consisting of malaria T-cell epitope, an outer membrane protein of Neisseria Meningitidis, cationized bovine serum albumin, Keyhole Limpet Hemocyanin, polylysine and human serum albumin; and

wherein in the conjugate the ganglioside derivative is covalently bound to the immunogenic protein-based carrier by a stable amine bond between the C-4 carbon of the altered sphingosine base of the altered ceramide portion of the ganglioside derivative and the nitrogen of an ϵ -aminolysyl group of the immunogenic protein-based carrier.--

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--124. (New) The composition of claim 123, wherein the saponin is QS-21.--

--125. (New) The composition of claim 123, wherein the amount of the conjugate is an amount of between about 1 μg and about 200 μg .--

--126. (New) The composition of claim 125, wherein the amount of the conjugate is an amount of between 10 μg and 90 μg .--

--127. (New) The composition of claim 125, wherein the amount of the conjugate is an amount of between 10 μg and 70 μg . --

--128. (New) The composition of claim 125, wherein the amount of the conjugate is an amount of between 10 μg and 50 μg .--

 --129. (New) The composition of claim 123, wherein the amount of the saponin is an amount of between about 10 μg and about 200 μg .--

--130. (New) The composition of claim 129, wherein the amount of the saponin is about 100 μg .--

--131. (New) The composition of claim 129, wherein the amount of the saponin is about 200 μg .--

--132. (New) A composition which comprises:

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- a) a conjugate comprising (i) a ganglioside derivative which comprises an unaltered oligosaccharide part and an altered ceramide portion comprising an altered sphingosine base and (ii) an immunogenic protein-based carrier;
- b) a saponin derivable from the bark of a Quillaja saponaria Molina tree, wherein the saponin is QS-21; and
- c) a pharmaceutically acceptable carrier;

wherein the conjugate is present in an amount of between about 10 µg and about 50 µg, and the amount of the saponin is about 100 µg, and wherein the relative amounts of such conjugate and such saponin is effective to stimulate or enhance production in a subject of an antibody to GD2, GD3 and GT3, whichever ganglioside is present as a derivative in the conjugate,

wherein the ganglioside derivative is a derivative of a ganglioside selected from the group consisting of GD2, GD3 lactone, O-acetyl GD3 and GT3;

wherein the immunogenic protein-based carrier is derived from a protein selected from the group consisting of malaria T-cell epitope, an outer membrane protein of Neisseria Meningitidis, cationized bovine serum albumin, Keyhole Limpet Hemocyanin, polylysine and human serum albumin; and

wherein in the conjugate the ganglioside derivative is covalently bound to the immunogenic protein-based carrier by a stable amine

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bond between the C-4 carbon of the altered sphingosine base of the altered ceramide portion of the ganglioside derivative and the nitrogen of an ϵ -aminolysyl group of the immunogenic protein-based carrier.-

--133. (New) A method of treating a subject afflicted with melanoma which comprises administering to said subject an amount of a composition of claim 132 effective to stimulate or enhance production of an antibody to at least one ganglioside selected from the group consisting of GD2, GD3 lactone, O-acetyl GD3 and GT3 and to thereby treat said melanoma in said subject.--

II
--134. (New) A method of stimulating or enhancing production of an antibody to a GD2, GD3 and GT3 in a subject which comprises administering to the subject an effective amount of a composition which comprises;

- a) a conjugate comprising (i) a ganglioside derivative which comprises an unaltered oligosaccharide part and an altered ceramide portion comprising an altered sphingosine base and (ii) an immunogenic protein-based carrier;
- b) a saponin derivable from the bark of a Quillaja saponaria Molina tree; and
- c) a pharmaceutically acceptable carrier;

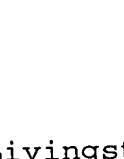
the relative amounts of such conjugate and such saponin being effective to stimulate or enhance production in a subject of an

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antibody to GD2, GD3 and GT3, whichever ganglioside is present as a derivative in the conjugate,

wherein the ganglioside derivative is a derivative of a ganglioside selected from the group consisting of GD2, GD3 lactone, O-acetyl GD3 and GT3;

wherein the immunogenic protein-based carrier is derived from a protein selected from the group consisting of malaria T-cell epitope, an outer membrane protein of Neisseria Meningitidis, cationized bovine serum albumin, Keyhole Limpet Hemocyanin, polylysine and human serum albumin; and

 wherein in the conjugate the ganglioside derivative is covalently bound to the immunogenic protein-based carrier by a stable amine bond between the C-4 carbon of the altered sphingosine base of the altered ceramide portion of the ganglioside derivative and the nitrogen of an ϵ -aminolysyl group of the immunogenic protein-based carrier, so as to thereby stimulate or enhance production of the antibody to GD2, GD3 and GT3 in the subject, whichever ganglioside is present as a derivative in the conjugate.--

--135. (New) A method of treating a cancer in a subject which comprises administering to the subject an effective cancer-treating amount of a composition which comprises:

- a) a conjugate comprising (i) a ganglioside derivative which comprises an unaltered oligosaccharide part and an altered ceramide portion comprising an altered

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sphingosine base and (ii) an immunogenic protein-based carrier;

b) a saponin derivable from the bark of a Quillaja saponaria Molina tree; and

c) a pharmaceutically acceptable carrier;

the relative amounts of such conjugate and such saponin being effective to stimulate or enhance production in a subject of an antibody to GD2, GD3 and GT3, whichever ganglioside is present as a derivative in the conjugate;

wherein the ganglioside derivative is a derivative of a ganglioside selected from the group consisting of GD2, GD3 lactone, O-acetyl GD3 and GT3;

wherein the immunogenic protein-based carrier is derived from a protein selected from the group consisting of malaria T-cell epitope, an outer membrane protein of Neisseria Meningitidis, cationized bovine serum albumin, Keyhole Limpet Hemocyanin, polylysine and human serum albumin; and

wherein in the conjugate the ganglioside derivative is covalently bound to the immunogenic protein-based carrier by a stable amine bond between the C-4 carbon of the altered sphingosine base of the altered ceramide portion of the ganglioside derivative and the nitrogen of an ϵ -aminolysyl group of the immunogenic protein-based carrier, so as to thereby stimulate or enhance production of the antibody to GD2, GD3 and GT3 in the subject, whichever

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ganglioside is present as a derivative in the conjugate.--

--136. (New) The method of claim 135, wherein the cancer is of epithelial origin.--

--137. (New) The method of claim 135, wherein the cancer is of neuroectodermal origin.--

--138. (New) The method of claim 137, wherein the cancer of neuroectodermal origin is a melanoma.--

--139. (New) The method of claim 134 or 135, wherein the administering is effected at two or more sites.--

--140. (New) The method of claim 139, wherein the administering is effected at three sites.--

--141. (New) The method of claim 134 or 135, wherein the composition is administered subcutaneously to said subject.--

--142. (New) The method of claim 141, wherein the composition is administered to said subject at two-week intervals.--

--143. (New) The method of claim 141, wherein the composition is administered to said subject at weekly intervals.--

--144. (New) The method of claim 134 or 135, wherein the composition to be administered is prepared prior to administration to the subject by mixing the conjugate and the saponin.--

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--145. (New) The method of claim 144, wherein the conjugate and the saponin are mixed on the day of administration to the subject.--

--146. (New) A method of delaying recurrence of melanoma in subjects at risk of relapse of melanoma which comprises administering to the subject an effective amount of a composition which comprises:

- I
- a) a conjugate comprising (i) a ganglioside derivative which comprises an unaltered oligosaccharide part and an altered ceramide portion comprising an altered sphingosine base and (ii) an immunogenic protein-based carrier;
 - b) a saponin derivable from the bark of a Quillaja saponaria Molina tree; and
 - c) a pharmaceutically acceptable carrier;

the relative amounts of such conjugate and such saponin being effective to stimulate production in a subject of an antibody to GD2, GD3 and GT3, whichever ganglioside is present as a derivative in the conjugate;

wherein the ganglioside derivative is a derivative of a ganglioside selected from the group consisting of GD2, GD3 lactone, O-acetyl GD3 and GT3;